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DEPARTMENT OF THE ARMY
Fort Detrick
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Sensitivity Test in vivo of Brucella to Antibiotics

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In preceding experiments we have attempted to use the behavior of the focal site determined from inoculation of brucella in rabbit skin as a test of sensitivity in vivo to chloramphenicol (1)

The results of this experiment were not satisfactory, which posed the question of whether the inefficacy of treatment was due to non-validity of the method used, or whether it should be attributed to the limited activity of the medication. For the solution of such a problem, we have studied comparatively the activity of chloramphenicol, chlorotetracycline and neomycin on intradermal infection, effected first with a dose of Br. melitensis (16a) in a single dermal site and in multiple dermal sites, and in a succeeding experiment with 4 doses inoculated contemporaneously in such manner as to have comparative evaluations of their sensitivity. The doses used are: Br. melitensis 16a, Br. suis 1330, Br. abortus 544 and 99 W, this last aerobic and of limited virulence. The two experiments were done in 14 rabbits.

For treatment the following antibiotics were used: chlorotetracycline and chloramphenicol in aqueous suspension 5%, neomycin in aqueous solution 10%. The provision of antibiotics was made intramuscularly in single doses daily from g 0,1.

Sensitivity in vitro to antibiotics has been tested in Albimi broth, with dilutions of antibiotics growing in geometric progression of tens. The development of the lesions in the animals is followed, taking into account the diameter, the entity of infiltration and erythema, and

eventually the appearance at necropsy, of scab and ulcer.

After 12 days the animals are sacrificed and isolations made from the sites of inoculation (potato agar) from the liver and from the spleen. Periodically (4th, 8th, 12th days) the rate of agglutininis controlled.

In the first experiment (infection with 16m; 1 x 4 sites), no difference is observed in the behavior of the lesion between rabbits with one single site and rabbits with multiple sites, contrary to what seemed suggested in a preceding experiment with impiego of CAF (1). The single components of the infection (erythema, infiltrated) were notably reduced by treatment with neomycin and to a lesser extent by treatment with chlorotetraciline. Treatment with neomycin prevented evolution of focal site toward necrosis. Lesions of animals treated with CAF were modestly reduced in the first days of treatment, consequently have taken a course quite parallel to that of the control.

Production of agglutinating antibodies has been more modest in animals with a single site of inoculation, evidently in accordance with the less stimulating antigen, but was not influenced by treatment with antibiotics.

Positive isolations were had from dermal sites of rabbits treated with chlorotetraciline, from the spleen, from the liver, and from dermal sites of rabbits treated with CAF and from control rabbits; on the contrary isolations, whether from the organs or from dermal sites of rabbits treated with neomycin are always negative. Those reisolated have not shown any variation in sensitivity in vitro to antibiotics (sensitivity from 1 - 10 y).

The results of this experiment have been confirmed by succeeding experiment with impiego of various doses: there was observed a little difference of sensitivity in vivo between those assayed: more sensitive were Br. suis 1330 and Br. abortus 99W. At the same time there was tried also inoculation with a mixture of brucella with antibiotic, added in doses

assuredly inhibiting in vitro. Such mixtures have been inoculated by intradermal route immediately after their preparation, with a practically negligible contact in vitro.

Lesion is not manifested in the rabbit inoculated with brucella and neomycin, and is notably reduced in the rabbit inoculated with brucella and chlorotetracycline and only retarded in the rabbit inoculated with brucella and CAF. The rate of agglutinin is lower in the rabbit treated with the suspension of brucella and neomycin, in relation to a reduced stimulating antigen, the focal site not being seen.

Isolations from the organs are shown positive in animals treated with CAF, in the rabbit infected with brucella and chlorotetracycline and in the controls, negative in rabbits treated with chlorotetracycline by general route and in rabbits treated with neomycin whether by general route or local.

The results are evidently parallel between the evolution of the dermal lesion and the diffusion of the infection.
The results are evidently parallel between the evolution of the dermal lesion and the diffusion of the infection. Therapy with neomycin or chlorotetracycline shows efficacy on the local site, impedes generalization of the infection; treatment with CAF on the other hand does not modify the evolution of the local site and does not hinder generalization of infection. () ←

The results seem to us indicative of two different possibilities of development: attempts at antibiotic therapy, treating infected animals by intradermal route, with medication generalized; study of activity of antibiotics effecting titrations in their neutralizing capacity on dermopathogenic activity of brucella.

(1) Gargani G. and Lo Monaco G. B., N. Ann. Igiene e Microbiologia, 1955, 6.